

AMENDMENTS TO THE CLAIMS

Claim 1 (currently amended): A method of determining total urokinase concentration in a sample containing ~~either or both~~ at least one of an active and or inactive form[[s]] of urokinase, comprising:

~~obtaining at least one peptide comprised of:~~

~~a first peptide corresponding to a sequence of SEQ ID No. 16 between amino acid residues 1 and 135,~~

~~a second peptide corresponding to a sequence of SEQ ID No. 16 between amino acid residues 159 and 411,~~

~~a third peptide corresponding to a sequence of SEQ ID No. 16 which includes amino acid residues 158 and 159, or~~

~~any combination;~~

~~obtaining at least one immunological composition directed against each of said at least one peptide;~~

~~contacting aliquots of said sample with each of said at least one immunological composition;~~

~~determining total urokinase concentration in said sample by determining the quantity of each of said at least one immunological composition that is bound to at least one of said forms of urokinase in each of said aliquots.~~

generating at least one immunological composition directed against at least one of a first peptide with SEQ ID NOS: 1, 2, 3, 4, 5, 6, at least one of a second peptide with SEQ ID NOS: 7, 8, 9, 10, 11 or 12 and at least one of a third peptide

with SEQ ID NOs: 13 or 14 or functionally equivalent peptide(s) containing amino acid substitution(s) with a difference in the hydropathic index value of $\pm 1-2$ from said corresponding first peptide, second peptide or third peptide;

contacting said sample with each of said at least one immunological composition;

measuring quantity and comparing type of said at least one immunological composition bound in said sample to determine a concentration of said at least one of said active or inactive form of urokinase, wherein each of said at least one immunological composition binds to said at least one of said active or inactive form of urokinase and is indicative of said concentration of the form that is bound thereto, wherein the total of said concentration of said at least one of said active and inactive form of urokinase represents the total urokinase concentration in said sample.

Claims 2-4. (canceled).

Claim 5 (currently amended): The method of claim 1, wherein each of said at least one immunological composition has a binding affinity constant for said ~~at least one first peptide, said second peptide or said third peptide~~ from against which it is directed ~~against~~ that is substantially higher than its binding affinity constant for a non-urokinase protein as similar in amino acid sequence to urokinase as is trypsin.

Claim 6 (currently amended): The method of claim 1, wherein ~~said third peptide corresponds to Seq ID No. 14, wherein said step of obtaining an immunological composition directed against each of said at least one peptide is used to obtain an immunological composition directed against said third peptide corresponding to Seq ID No. 14, and wherein~~ said at least one immunological composition directed against ~~said a~~ third peptide with ~~corresponding to Seq~~ SEQ ID NO: [[No.]] 14 exhibits a binding affinity constant for urokinase zymogen, an inactive form of urokinase, of at least 1×10^8 M⁻¹ and a binding affinity constant for forms of urokinase lacking a peptide bond between amino acid residues 158 and 159 of Seq ID No. 16 of at least approximately 10-fold lower than that for urokinase zymogen.

Claim 7 (canceled).

Claim 8 (canceled).

Claim 9 (currently amended): The method of claim 1, wherein said at least one immunological composition is an antiserum, an antibody, or a supernatant of a hybridoma, ~~or any combination~~ obtained via injection into a mammal of said at least one first peptide, second peptide and third peptide.

Claim 10 (currently amended): The method of claim 1, wherein said determination of said quantity of each of said at least one immunological composition is carried out by radiolabeling said sample ~~or said at least one immunological composition~~.

Claim 11 (currently amended): The method of claim 1, wherein ~~said determining said at least one active or inactive form of urokinase comprises total urokinase concentration in said sample by determining said quantity of each of said at least one immunological composition that is bound to at least one of said forms of urokinase in each of said aliquot is comprised of the following steps:~~

~~determining the a amount of low molecular weight urokinase in said sample that which is bound to binds to one or more of said at least one immunological composition directed against said second peptide corresponding to a sequence of Seq ID No. 16 between amino acid residues 159 and 411, but which does not bind is not bound to one or more of said at least one immunological composition directed against said first peptide; corresponding to a sequence in Seq ID No. 16 between amino acid residues 1 and 135;~~

~~determining the a amount of high molecular weight urokinase in said sample that which binds is bound to one or more of said at least one immunological composition directed against said first peptide or second peptide corresponding to a sequence in Seq ID No. 16 between amino acid residues 1 and 135 or between amino acid residues 159 and 411, respectively, but which does not bind is not bound to one or more of said at least one immunological composition directed against said third peptide;~~

~~and corresponding to a sequence in Seq ID No. 16 which includes amino acid residues 158 and 159~~

~~determining the a amount of urokinase zymogen, an inactive form of urokinase in said sample, that which binds is bound to one or more of said at least one immunological composition directed against said third peptide corresponding to a sequence of Seq ID No. 16 which includes amino acid residues 158 and 159, or any combination.~~

Claim 12 (canceled).

Claim 13 (currently amended): A method of determining total urokinase concentration in a sample containing ~~either or both~~ at least one of an active and or inactive form[[s]] of urokinase, comprising:

~~obtaining at least one peptide comprising:~~

~~a first peptide identical to a predetermined sequence in Seq ID No. 16 between amino acid residues 1 and 135,~~

~~a second peptide identical to a predetermined sequence in Seq ID No. 16 between amino acid residues 159 and 414~~

~~a third peptide identical to a predetermined sequence in Seq ID No. 16 which includes amino acid residues 158 and 159, or~~

~~any combination;~~

~~obtaining~~ generating at least one immunological composition selected from ~~[[the]]~~ a group of immunological compositions consisting of an antisera, an antibody and a supernatant of a hybridoma, said at least one immunological composition each obtained via injection ~~into a mammal~~ of said at least one of a first peptide with SEQ ID NOs: 1, 2, 3, 4, 5 or 6, at least one of a second peptide with SEQ ID NOs: 7, 8, 9, 10, 11 or 12 and at least one of a third peptide with SEQ ID NOs: 13 or 14 or functionally equivalent peptide(s) containing amino acid substitutions with a difference in the hydropathic index value of $\pm 1-2$ from the corresponding first peptide, second peptide or third peptide;

contacting ~~aliquots of~~ said sample with each of said at least one immunological composition; and

~~determining the total urokinase concentration by determining said quantity of each of said at least one immunological composition that is bound to at least one of said forms of urokinase in each of said aliquots, comprising the following steps:~~

measuring quantity and comparing type of each of said at least one immunological composition bound in said sample to determine a total concentration of said at least one of said active and inactive form of urokinase in said sample, wherein each of said at least one immunological composition binds to said at least one of said active or inactive forms of urokinase and is indicative of said total concentration of the form of urokinase that is bound thereto, said determination comprising:

determining the amount a first concentration of low molecular weight urokinase in said sample, wherein said low molecular weight urokinase which binds is

bound to ~~one or more of~~ said at least one immunological composition directed against said second peptide ~~identical to any of Seq ID Nos. 7-12, but which does not bind~~ is not bound to ~~one or more of~~ said at least one immunological composition directed against said first peptide; ~~identical to any of Seq ID Nos. 1-6,~~

determining the amount a second concentration of high molecular weight urokinase in said sample, wherein said high molecular weight urokinase which binds is bound to ~~one or more of~~ said at least one immunological composition directed against said first peptide or second peptide ~~identical to any of Seq ID Nos. 1-6 or Seq ID Nos. 7-12, respectively, but which does not bind~~ is not bound to ~~one or more of~~ said at least one immunological composition directed against said third peptide; ~~identical to Seq ID Nos. 13 or 14,~~

determining the amount a third concentration of urokinase zymogen, an inactive form of urokinase, in said sample, wherein said urokinase zymogen which binds is bound to ~~one or more of~~ said at least one immunological composition directed against said third peptide; and identical to Seq ID Nos. 13 or 14, or

any combination, adding the first, second, and third concentrations to obtain said total concentration, wherein said total concentration represents a total urokinase concentration in said sample.

Claim 14. (currently amended): A kit for determining total urokinase concentration in a sample containing ~~either or both~~ at least one of an active and or inactive form[[s]] of urokinase, comprising:

~~at least one immunological composition(s) wherein said at least one immunological composition is each obtained via injection into a mammal of at least one peptide, wherein each of said at least one immunological composition is directed against one or more of said at least one peptide, and wherein said at least one peptide is comprised of:~~

~~a first peptide identical to a predetermined sequence in Seq ID No. 16 between amino acid residues 1 and 135,~~

~~a second peptide identical to a predetermined sequence in Seq ID No. 16 between amino acid residues 159 and 411,~~

~~a third peptide identical to a predetermined sequence in Seq ID No. 16 which includes amino acid residues 158 and 159, or~~

~~any combination; immunological composition(s) directed against at least one of a first peptide with SEQ ID NOs: 1, 2, 3, 4, 5, or 6, at least one of a second peptide with SEQ ID NOs: 7, 8, 9, 10, 11 or 12 and at least one of a third peptide with SEQ ID NOs: 13 or 14 or against functionally equivalent peptide(s) containing amino acid substitution(s) with a difference in the hydrophatic index value of ± 1 to 2 from the corresponding first peptide, second peptide or third peptide; and~~

~~instructions for contacting aliquots of said sample with each of said at least one immunological composition and determining said total urokinase concentration in said sample by adding concentrations of said at least one of said active and inactive form of urokinase obtained by measuring quantity and type of immunological composition(s) bound in said sample, determining the quantity of each of said at least one~~

~~immunological composition which is bound to at least one of said forms of urokinase in each of said aliquots.~~

Claims 15-18 (canceled).

Claim 19 (currently amended): The kit of claim 14, ~~wherein said at least one peptide~~ wherein said immunological composition(s) is further comprised of:

an immunological composition directed against a fourth peptide with SEQ ID NO: 17 or against functionally equivalent peptide(s) containing amino acid substitution(s) with a difference in the hydropathic index value of ± 1 to 2 from the corresponding fourth peptide.

~~comprised of a fourth peptide identical to Seq ID No. 17.~~

Claim 20 (currently amended): The kit of claim 14, wherein said ~~at least one~~ immunological composition(s) is an antiserum, an antibody[[,]] or a supernatant of a hybridoma, ~~or any combination.~~

Claim 21 (canceled).

Claim 22 (withdrawn): A peptide for determining total urokinase concentration in a sample as in any of Seq ID Nos. 1-15 and 17.

Claim 23 (withdrawn): An antisera obtained via injection into a mammal of a peptide for determining total urokinase concentration in a sample as in any of Seq ID Nos. 1-15 and 17.

Claim 24 (withdrawn): An antibody obtained via injection into a mammal of a peptide for determining total urokinase concentration in a sample as in any of SEQ ID Nos. 1-15 and 17.

Claim 25 (withdrawn): A hybridoma producing an antibody obtained via injection into a mammal of a peptide for determining total urokinase concentration in a sample as in any of Seq ID Nos. 1-15 and 17.

Claim 26 (withdrawn): A supernatant of a hybridoma producing an antibody obtained via injection into a mammal of a peptide for determining total urokinase concentration in a sample as in any of Seq ID Nos. 1-15 and 17.

Claim 27 (new): The method of claim 1, wherein said step of generating at least one immunological composition is further comprised of generating at least one immunological composition directed against a fourth peptide with SEQ ID NO: 17 or functionally equivalent peptide(s) containing amino acid substitutions with a difference in the hydropathic index value of $\pm 1-2$ from said fourth peptide.

Claim 28 (new): The method of claim 27, wherein said at least one of said active or inactive form of urokinase comprises a low molecular weight urokinase that is bound to said at least one of said immunological composition directed against said second peptide, but is not bound to said at least one immunological composition directed against said fourth peptide; and a high molecular weight urokinase in the sample that is bound to said at least one immunological composition directed against said first peptide or said second peptide, but is not bound to said at least one immunological composition directed against said third peptide.